Proposal form for the evaluation of a genetic test for NHS Service Gene Dossier/Additional Provider

TEST – DISORDER/CONDITION – POPULATION TRIAD			
Submitting laboratory:	**Bristol RGC**	**Approved:**	**Sept 2013**
***1. Disorder/condition – approved name and symbol as published on the OMIM database** (alternative names will be listed on the UKGTN website)	EPISODIC KINESIGENIC DYSKINESIA 1; EKD1 SEIZURES, BENIGN FAMILIAL INFANTILE, 2; BFIS2 CONVULSIONS, FAMILIAL INFANTILE, WITH PAROXYSMAL CHOREOATHETOSIS; ICCA		
2. OMIM number for disorder/condition	128200, 605751, 602066		
3a. Disorder/condition – please provide, in laymen's terms, a brief (2-5 sentences) description of how the disorder(s) affect individuals and prognosis.	These allelic disorders are autosomal dominant neurological conditions and can be regarded as clinical variations of the same syndrome. This is an emerging common group of paroxysmal movement disorders. They are characterised by afebrile seizures and various degrees of dystonia. Familial Paroxysmal Kinesigenic Dyskinesia (EKD1) has onset in childhood or adolescence, while in Benign familial Infantile Convulsions (BFIS) and Benign familial Infantile Convulsions with paroxysmal choreoathetosis (ICCA) the average onset of seizures is 6 months. Symptoms become less severe with age and show favourable response to anticonvulsant medications such as carbamazepine or phenytoin. The condition is often difficult to distinguish from other forms of epilepsy.		
3b Disorder/condition – if required please expand on the description of the disorder provided in answer to Q3a.			
4. Disorder/condition – mode of inheritance	Autosomal dominant		
5. Gene – approved name(s) and symbol as published on HGNC database (alternative names will be listed on the UKGTN website)	Proline-rich transmembrane protein 2; PRRT2		
6a. OMIM number for gene(s) **6b HGNC number for gene(s)**	OMIM 614386 HGNC 30500		
7a. Gene – description(s)	The PRRT2 gene is located on chromosome 16p11.2, it is consisted of 4 exons, the first one is not coding. The protein encoded by *PRRT2* (NP_660282.2) has 340 amino acids and is predicted to have two transmembrane segments. The function is unknown; however, yeast two-hybrid studies suggest that PRRT2 interacts with synaptosomal-associated protein 25kd (SNAP25). High levels of PRRT2 mRNA have been identified in the globus pallidus, cerebellum, subthalamic nucleus, cerebellar peduncles, caudate nucleus, and cerebral cortex. The truncated PRRT2 protein results in altered subcellular localization.		
7b. Number of amplicons to provide this test (molecular) or type of test (cytogenetic)	4 amplicons (3 for exon 2 and 1 for exons 3 and 4).		

7c. GenU band that this test is assigned to for index case testing	Band C
8. Mutational spectrum for which you test including details of known common mutations	To date more than 20 mutations have been described in the literature; these include missense, frameshift and splice site mutations. Mutations are distributed throughout the gene; there are no reported mutations in non-coding exon 1. The c.649dupC is the most common PRRT2 mutation reported to date. It is located in exon 2 and results in a frameshift with the insertion of a premature stop codon (p.Arg217Profs*8). The c.649dupC is a recurrent frameshift, loss of function mutation, which has been identified in families and in sporadic patients with paroxysmal kinesigenic dyskinesia, with or without infantile convulsions and benign familial infantile epilepsy (Marini et al, 2012). PRRT2 mutations have a high prevalence in patients with benign familial infantile seizures either alone or in association with paroxysmal kinesigenic or exercise-induced dyskinesia (40-90%), but are only rarely detected in sporadic cases (Guerrini et al, 2012). Mutation rates in familial autosomal dominant paroxysmal kinesigenic dyskinesia is about 89% and falls to 50% when only index cases are considered (Guerrini et al, 2012). Heron et al. reported PRRT2 mutations in 82% of BFIS affected patients and 83% in ICCA patients. The c.649dupC is the most common recurrent and frequent PRRT2 mutation reported to date. Heron et al. (2012) reported the 649dupC (p.Arg217fs) mutation in 79% of families with ICCA or BFIS. Schubert et al. reported an overall pick up rate of 83%; with 77% for the BFIS patients carrying the common c.649dupC mutation. Marini et al. Neurology 2012; 79:2109-2114 Guerrini et al. Neurology 2012; 79:2086-2088 Heron et al. Am J Hum Genet 2012; 90:152-160 Schubert et. Hum Mutat 2012; 33(10): 1439-1443
9a. Technical method(s)	High Throughput (HT) Automated Sequence Analysis. Gene screening by bidirectional automated Sanger sequence analysis (BiomekNX and ABI3730), the standard platform used for gene screening at BGL and analysis of the results with Mutation Surveyor software.
9b If a panel test using NGS please state if it is a conventional panel or a targeted exome test.	n/a
9c. Panel/targeted exome Tests **i) Do the genes have 100% coverage? If not what is the strategy for dealing with the gaps in coverage?**	n/a

ii) Does the test include MLPA?	
iii) Does this use sanger sequencing or Next Generation Sequencing (NGS)?	
iv) If NGS is used, does the lab adhere to the Practice Guidelines for NGS?	
10 Is the assay to be provided by the lab or is it to be outsourced to another provider? **If to be outsourced, please provide the name of the laboratory.**	It is provided by this laboratory (BGL).
11. Validation process Please explain how this test has been validated for use in your laboratory or submit your internal validation documentation	HT automated sequence analysis: This method is routinely used to screen for mutations at BGL and had been validated for many services (number of amplicons currently in service: 304). Beckman NX robotics /ABI 3730 capillary electrophoresis and mutation surveyor software analysis are routinely used in the laboratory. Positive and negative control samples are included. Primers (SNP checked) are designed to cover the coding regions and intron-exon boundaries. BGL also participates in external quality assurance EMQN sequencing QA schemes (since the pilot scheme was introduced in 2002) and UV interpretation scheme (pilot scheme introduced in 2012).
12a. Are you providing this test already?	☐ **No** ☒ **Yes**
12b. If yes, how many reports have you produced? Please provide the time period in which these reports have been produced and whether in a research or a full clinical diagnostic setting.	One
12c. Number of reports mutation positive	One
12d. Number of reports mutation negative	
13. For how long have you been providing this service?	Since December 2012
14a. Is there specialised local clinical/research expertise for this disorder?	☐ **No** ☒ **Yes**
14b. If yes, please provide details	Dr Philip Jardine Consultant Paediatric Neurologist Dr Sarah Smithson Consultant Clinical Geneticist
15. Are you testing for other genes/disorders/conditions closely allied to this one? Please give details	No
16. Based on experience what will be the national (UK wide) activity, per annum, for:	
16a. Index cases	18
16b. Family members where mutation is known	25

17a. Does the laboratory have capacity to provide the expected national activity?	Yes
17b. If your laboratory does not have capacity to provide the full national need please could you provide information on how the national requirement may be met. For example, are you aware of any other labs (UKGTN members or otherwise) offering this test to NHS patients on a local area basis only? This question has been included in order to gauge if there could be any issues in equity of access for NHS patients. It is appreciated that some laboratories may not be able to answer this question. If this is the case please write "unknown".	
18. Please justify the requirement for another laboratory to provide this test e.g. insufficient national capacity.	

EPIDEMIOLOGY

19a. Estimated prevalence of condition in the general UK population	The prevalence of Familial Paroxysmal Kinesigenic Dyskinesia is estimated at 1:150,000 (van Rootselaar et al, 2009) The prevalence for Benign Familial Infantile Convulsions with or without paroxysmal choreoathetosis is currently unknown. Fewer cases are described in the literature, but this may change with increasing awareness. van Rootselaar et al. Pract Neurol 2009;9:102-109
19b. Estimated incidence of condition in the general UK population Please identify the information on which this is based	There are no epidemiological studies to give an accurate incidence of the condition.
20. Estimated gene frequency (Carrier frequency or allele frequency) Please identify the information on which this is based	No evidence is available in the literature.
21. Estimated penetrance Please identify the information on which this is based	There is variable penetrance, ranging from 60 to 90% depending on the clinical context. The penetrance of PRRT2 mutations is approximately 60%, if only the Paroxysmal Kinesigenic Dyskinesia (PKD) phenotype is considered; but if infantile convulsions are also taken into account, the penetrance is 90% (van Vliet et al, 2012). Another study has reported the penetrance for Familial Paroxysmal Kinesigenic Dyskinesia and Benign familial Infantile Convulsions with paroxysmal Choreoathetosis as approximately 80% (Heron et al, 2012) van Vliet et al. Neurology 2012;79:777-784 Heron et al. Am J Hum Genet 2012; 90:152-160
22. Estimated prevalence of condition in the population of people that meet the Testing Criteria.	Not known.

INTENDED USE

23. Please tick either yes or no for each clinical purpose listed.

Panel Tests: a panel test would not be used for pre symptomatic testing, carrier testing and pre natal testing as the familial mutation would already be known in this case and the full panel would not be required.

Diagnosis	☒ **Yes**	☐ **No**
Treatment	☒ **Yes**	☐ **No**
Prognosis & management	☒ **Yes**	☐ **No**
Presymptomatic testing (n/a for panel tests)	☐ **Yes**	☒ **No**
Carrier testing for family members (n/a for panel tests)	☒ **Yes**	☐ **No**
Prenatal testing (n/a for panel tests)	☒ **Yes on a case by case basis** ☐ **No**	

TEST CHARACTERISTICS

24. Analytical sensitivity and specificity

This should be based on your own laboratory data for the specific test being applied for or the analytical sensitivity and specificity of the method/technique to be used in the case of a test yet to be set up.

High Throughput Semi Automated Sequence analysis.
Sensitivity 99-100%.
To the best of our knowledge, no variant has been missed using a bi-directional sequencing approach.
Current validation of unidirectional sequencing within SCOBEC indicates a sensitivity of 99%.

Specificity > 99%

25. Clinical sensitivity and specificity of test in target population

The *clinical sensitivity* of a test is the probability of a positive test result when condition is known to be present; the *clinical specificity* is the probability of a negative test result when disorder is known to be absent. The denominator in this case is the number with the disorder (for sensitivity) or the number without condition (for specificity).

The sensitivity for detecting a mutation in PRRT2 when it is the gene involved will be close to 100% as all the mutations described so far are point mutations. The sensitivity in patients who have a diagnosis of one of these allelic disorders will be lower than 100% as heterogeneity is observed:

in different studies the PRRT2 mutations account for > 80% of BFIS [1,2,3], 83% of ICCA cases [1],
[1] Heron et al. Am J Hum Genet 2012; 90:152-160
[2] Schubert et. Hum Mutat 2012; 33(10): 1439-1443
[3] Scheffer et al Neurology 2012; 79: 2104-2108

With variable penetrance, and the possibility of mutational uncertainty due to novel uncharacterised mutations, realistic figures would be close to 100% for sensitivity and probably 90% for specificity.

For testing of at-risk relatives: Clinical sensitivity will be 100%
Clinical sensitivity and specificity: will both be 100% if the mutation is definitely pathogenic and the disease is fully penetrant. With possible reduced penetrance, and the possibility of mutational uncertainty, realistic figures would be close to 100% for sensitivity and probably 90% for specificity where defined as future clinical manifestation.

26. Clinical validity (positive and negative predictive value in the target population)

The *clinical validity* of a genetic test is a measure of how well the test predicts the presence or absence of the phenotype, clinical condition or predisposition. It is measured by its *positive predictive value* (the probability of getting the condition given a positive test) and *negative predictive value* (the probability of not getting the condition given a negative test).

Clinical validity: In an index case, finding a pathogenic mutation confirms the diagnosis; absence of a mutation does not exclude the diagnosis.

For index cases:
Positive predictive value (PPV) = 100% for consensus mutations

Negative predictive value (NPV) = close to 100% if defining the disease, but given the heterogeneity observed, in practice NPV will be <100%.

However, for testing family members, PPV and NPV are both effectively 100% for predisposition for symptoms using consensus mutations.

27. Testing pathway for tests where more than one gene is to be tested

Please include your testing strategy if more than one gene will be tested and data on the expected proportions of positive results for each part of the process. Please illustrate this with a flow diagram. This will be added to the published Testing Criteria.

n/a

CLINICAL UTILITY

28. How will the test change the management of the patient and/or alter clinical outcome?

For BFIS and ICCA the test will be clinically useful to inform the clinical management of very young children, especially the prognosis of the seizures. Early-onset seizures have a wide variety of causes in this age group and some conditions are associated with a very poor outcome.

The age specific incidence of seizures is higher in infancy than at almost any other age (Olafsson et al. 2006). The majority of seizures in infancy are symptomatic (Eltze et al. Epilepsia 2012) and therefore extensive investigation is often necessary to elucidate the underlying aetiology. The prognosis associated with many forms of infantile seizures is poor in relation to both seizure control and neurodevelopmental outcome but the prognosis is often not clear at the onset of symptoms and before identification of aetiology. Identification of a PRRT2 mutation known to be associated with BFIS / ICCA is likely to result in a significantly reduced investigation burden and may increase the threshold for initiation of anti-epileptic drug therapy. Choice of AED may also be influenced by identification of a PRRT2 mutation as carbamazepine is known to be very effective in PKD (although the literature on treatment of BFIS / ICCA is very sparse).

Symptomatic cases of PKD are also known to occur (Strzelczyk et al. 2011) and identification of a PRRT2 mutation would negate the requirement to investigate for other aetiologies.

Olafsson et al. Lancet Neurol 2006;4:627-634

Eltze et al. Epilepsia 2012 - In print

Strzelczyk et al. 2011 Expert Opinion Pharmacotherapy 2011;12:63-72

29. Benefits of the test for the patient & other family members

Please provide a summary of the overall benefits of this test.

Positive molecular genetic diagnosis will direct treatment with carbamazepine which is usually highly effective.

Use of molecular genetics testing will reduce misdiagnosis and delayed diagnosis of PKD; a fact well described in the literature (van Rootselaar et al. Pract Neurol 2009;9:102-109).

Making these diagnoses is very reassuring for patients as more severe disorders can be excluded. Early diagnosis can avoid more invasive and expensive tests as well as hospital admissions to facilitate investigation.

30. What will be the consequences for patients and family members if this test is not approved?

On-going uncertainty about the cause of the seizures and confusion with other disorders. In older children the diagnosis of EKD1 may easily be missed or attributed to other problems rather than being recognised as a genetic condition. This misunderstanding may affect a child's general well-being and education.

At present no other positive diagnostic findings support the diagnosis of PKD / BFIS / ICCA and, they are to some extent "diagnoses of exclusion". Without molecular genetic testing on-going uncertainty about the diagnosis may lead to continued investigation or may lead to a more protracted treatment course than necessary.

31. Is there an alternative means of diagnosis or prediction that does not involve molecular diagnosis? If so (and in particular if there is a biochemical test), please state the added advantage of the molecular test.

As stated in the response to question 30, PKD / BFIS / ICCA are "diagnoses of exclusion"; clinical diagnosis may be supported by normal metabolic tests and normal neuroimaging.

32. Please describe any specific ethical, legal or social issues with this particular test.

No ethical issues are envisaged but advantages are possible as described above.

Because familial PKD demonstrates incomplete penetrance, a clinically unaffected parent can still have a *PRRT2* mutation.

This will have implications for the sibs of the proband, as there will be at a 50% risk of inheriting the mutation.

It is also useful to clinically evaluate an apparently asymptomatic parent with a thorough history and neurologic examination.

A positive genetic result on an asymptomatic parent can be followed by assessing the risk for other family members, for example his/her siblings, as they may also be asymptomatic and have the mutation, which similarly presents a risk to their offspring.

33. Only complete this question if there is previously approved Testing Criteria and you do not agree with it.

Please provide revised Testing Criteria on the Testing Criteria form and explain here the changes and the reasons for the changes.

34. List the diagnostic tests/procedures that an index case no longer needs if this genetic test is available.

Multiple blood tests
Repeated EEGs or 24 hour-EEG
Lumbar puncture
ECG
Hospital admissions for investigations
Brain scan may not needed in all cases if genetic testing result is available quickly; however as BFIS cases have been reported to have focal seizures, there may be cases that urgent imaging will be required.

In most cases the procedures mentioned above will take place in at least two different occasions, as separate admissions; however since there may still be some need of some of these tests, in the cost calculation only one day of hospital admission has been included.
-Intracranial Procedures Except Trauma with Muscular, Balance, Cranial or Peripheral Nerve disorders or Epilepsy- category 1 or 2 One day case HRG-£1957

	Type of test	**Cost (£)**
Costs and type of imaging procedures	MRI, CT, EEG, ECG, lumbar puncture/ admission in hospital **once**	£1957
Costs and types of laboratory pathology tests (other than molecular/cyto genetic test proposed in this gene dossier)	Blood tests	£500
Costs and types of physiological tests (e.g. ECG)	ECG	Included above
Cost and types of other investigations/procedures (e.g. biopsy)	Lumbar puncture	Included above
Total cost tests/procedures no longer required	**£**	**£2,457**

35. Based on the expected annual activity of index cases (Q15a), please calculate the estimated annual savings/investments based on information provided in Q33.

Number of index cases expected annually	(a) 18
Cost to provide tests for index cases if the genetic test in this gene dossier was not available (see Q34)	(b) £2,457
Total annual costs pre genetic test	(a) x (b) = (c) £44,226
Total annual costs to provide genetic test	(a) x cost of genetic testing for index case = (d) £5400
Total savings	(c) – (d) 44226 – 5400 = £38,826 saving

36. REAL LIFE CASE STUDY
In collaboration with the clinical lead, describe TWO real case examples:
1. **prior to availability of genetic test**
2. **post availability of genetic test**

to illustrate how the test improves patient experience and the costs involved.

Case example one – pre genetic test

A 5 month old boy born at term and previously well presented to the emergency department following a right-sided focal seizure that then became secondary generalised. The whole seizure lasted 4 minutes. The mother of the child had also had seizures as a child, but did not know any other details.

The child was making normal developmental progress and examination was normal. During the admission the child had two further short-lived seizures.

The child had a CT scan on admission. Multiple blood tests were sent for investigation of neuro-metabolic disorders. The child was started on sodium valproate. An MRI scan performed at a later date required a day case admission as general anaesthesia was required to perform the scan. A lumbar puncture was performed at the same time including an assay of CSF amino acids. A standard EEG was performed and as this was normal a prolonged ambulatory EEG was also performed. All investigations were normal but took 3 months for all results to be available. The child continued to make good developmental progress

PRE GENETIC TEST COSTS

	Type of test	Cost
Costs and type of imaging procedures	CT, MRI, EEGx2, Lumbar puncture	2x£1957
Costs and type of laboratory pathology tests	Blood tests	£500
Costs and type of physiological tests (e.g. ECG)		
Cost and type of other investigations/procedures (e.g. biopsy)		
Cost outpatient consultations (genetics and non genetics)	diagnostic x1 new case diagnostic x1 follow up Genetics x1 new	£396 £200 £1500
Total cost pre genetic test		**£6,510**

Case example two – post genetic test

An 8 month old girl who was previously well presented to the emergency department following a 3 minute generalised tonic-clonic seizure. By the time of admission she was back to normal. Developmental progress was normal and clinical examination was normal. The child's mother was reported to have had seizures as a young child that were treated with sodium valproate for 2-3 years. The mother has not had any problems since then. The maternal grandmother has migraine.

Routine blood tests were sent including basic screening for metabolic conditions. Blood for PRRT2

testing was also sent. The child was discharged and further investigations withheld pending the result of the PRRT2 testing.

POST GENETIC TEST COSTS

	Type of test	Cost
Costs and type of imaging procedures	MRI or EEG	£230
Costs and types laboratory pathology tests (other than molecular/cyto genetic proposed in this gene dosier)	Some blood tests	£250
Cost of genetic test proposing in this gene dossier	PRRT2 testing	£300
Costs and type of physiological tests (e.g. ECG)		
Cost and type of other investigations/procedures (e.g. biopsy)		
Cost outpatient consultations (genetics and non genetics)	diagnostic x1 new case diagnostic x1 follow up	£396 £200
Total cost post genetic test		**£1376**

UKGTN Testing Criteria

Test name: **Familial Paroxysmal Kinesigenic Dyskinesia and Benign Familial Infantile convulsions With or Without Choreoathetosis**	
Approved name and symbol of disorder/condition(s): Episodic Kinesigenic Dyskinesia 1; EKD1 Seizures, Benign Familial Infantile, 2; BFIS2 Convulsions, Familial Infantile, with Paroxysmal Choreoathetosis; ICCA	**OMIM numbers:** 128200, 605751, 602066
Approved name and symbol of gene(s): proline-rich transmembrane protein 2; PRRT2	**OMIM number:** 614386

Patient name:	**Date of birth:**
Patient postcode:	**NHS number:**
Name of referrer:	
Title/Position:	**Lab ID:**

Referrals will only be accepted from one of the following:	
Referrer	**Tick if this refers to you.**
Consultant Paediatric Neurologists	
Consultant Neurologists	
Consultant Clinical Geneticists	

Minimum criteria required for testing to be appropriate as stated in the Gene Dossier:	
Criteria	**Tick if this patient meets criteria**
Paroxysmal Kinesigenic Dyskinesia (PKD) - Paroxysmal movement disorder characterised by afebrile seizures and dystonia	
AND PKD - No loss of consciousness during episodes	
AND PKD - Normal neurological examination	
Benign Familial Infantile Convulsions(BFIS) - Onset of seizures in infancy (<2 years)	
AND BFIS - Normal neurodevelopment	
OR At risk family members where familial mutation is known.	

If the sample does not fulfil the clinical criteria or you are not one of the specified types of referrer and you still feel that testing should be performed please contact the laboratory to discuss testing of the sample.